

## PATENT COOPERATION TREATY

PCT

## NOTIFICATION OF ELECTION

(PCT Rule 61.2)

From the INTERNATIONAL BUREAU

To:

Assistant Commissioner for Patents  
 United States Patent and Trademark  
 Office  
 Box PCT  
 Washington, D.C.20231  
 ETATS-UNIS D'AMERIQUE

in its capacity as elected Office

<b>Date of mailing (day/month/year)</b> 11 August 2000 (11.08.00)	
<b>International application No.</b> PCT/EP99/09870	<b>Applicant's or agent's file reference</b> EA-PCT-10914
<b>International filing date (day/month/year)</b> 13 December 1999 (13.12.99)	<b>Priority date (day/month/year)</b> 14 December 1998 (14.12.98)
<b>Applicant</b> BACHER, Adelbert et al	

1. The designated Office is hereby notified of its election made:

☒ in the demand filed with the International Preliminary Examining Authority on:

14 July 2000 (14.07.00)

☐ in a notice effecting later election filed with the International Bureau on:2. The election ☒ was☐ was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO 34, chemin des Colombettes 1211 Geneva 20, Switzerland  Facsimile No.: (41-22) 740.14.35	Authorized officer  Pascal Piriou  Telephone No.: (41-22) 338.83.38
---	---

## PCT

## INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference <b>EA-PCT-10914</b>	<b>FOR FURTHER ACTION</b> see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.	
International application No. <b>PCT/EP 99/ 09870</b>	International filing date (day/month/year) <b>13/12/1999</b>	(Earliest) Priority Date (day/month/year) <b>14/12/1998</b>
Applicant <b>BACHER, Adelbert et al</b>		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 6 sheets.



It is also accompanied by a copy of each prior art document cited in this report.

## 1. Basis of the report

- a. With regard to the language, the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.



the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

- b. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of the sequence listing :



contained in the international application in written form.



filed together with the international application in computer readable form.



furnished subsequently to this Authority in written form.



furnished subsequently to this Authority in computer readable form.



the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.



the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

2. ☒ Certain claims were found unsearchable (See Box I).

3. ☐ Unity of invention is lacking (see Box II).

## 4. With regard to the title,



the text is approved as submitted by the applicant.



the text has been established by this Authority to read as follows:

## 5. With regard to the abstract,



the text is approved as submitted by the applicant.



the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

## 6. The figure of the drawings to be published with the abstract is Figure No.

3

as suggested by the applicant.



None of the figures.



because the applicant failed to suggest a figure.



because this figure better characterizes the invention.

# INTERNATIONAL SEARCH REPORT

International application No.

PCT/EP 99/09870

## Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:  
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 19-24

Present claims 19-24 relate to a compound defined by reference to a desirable property, namely its ability to inhibit at least one enzyme in the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate.

No technical features of the compounds are presented which would lead to this desirable property, the technical features formulated so as to permit the execution of a meaningful search. No means are identified by which agents known in the prior art could be distinguished from novel agents. For example, fosmidomycin (3-(N-formyl-N-hydroxyamino)propyl phosphonic acid), known in the prior art as a herbicide (see patent document EP256785, cited on the search report), and shown to behave as an inhibitor in the screening method of the present application (page 36), falls within the scope of claims 19-24. No definition of the subject matter for which protection is sought is therefore derivable from these claims (Article 6 PCT).

The claims cover all compounds having the property of inhibiting at least one enzyme in the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such compounds, which are in addition part of the prior art. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the compound by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

# INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 99/09870

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12Q1/02 G01N33/92 C07H13/00 C07H3/02 A01N61/00

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12Q G01N C07H

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>LANGE ET AL: "A family of transketolases that directs isoprenoid biosynthesis via a mevalonate-independent pathway"</p> <p>FASEB JOURNAL, US, FED. OF AMERICAN SOC. FOR EXPERIMENTAL BIOLOGY, BETHESDA, MD, vol. 95, March 1998 (1998-03), pages 2100-2104, XP002116672</p> <p>ISSN: 0892-6638</p> <p>page 2104, column 1, line 17, paragraph 2 - line 22</p> <p style="text-align: center;">--- -/--</p>	1-4, 18

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

\* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- "&" document member of the same patent family

Date of the actual completion of the international search

18 May 2000

Date of mailing of the international search report

07/06/2000

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  
Fax: (+31-70) 340-3016

Authorized officer

Hart-Davis, J

## International Application No.

C. (Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

1

## INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 99/09870

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,A	MCCASKILL, DAVID ET AL: "Isopentenyl diphosphate is the terminal product of the deoxyxylulose -5-phosphate pathway for terpenoid biosynthesis in plants" TETRAHEDRON LETT. (1999), 40(4), 653-656 , XP004151409 the whole document	
A	EP 0 256 785 A (FUJISAWA PHARMACEUTICAL CO) 24 February 1988 (1988-02-24) the whole document	
A	ROHMER, MICHEL: "Isoprenoid biosynthesis via the mevalonate - independent route, a novel target for antibacterial drugs?" PROG. DRUG RES. (1998), 50, 135-154 , XP000906878 the whole document	

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 99/09870

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
DE 19752700 A	02-06-1999	DE 29800547 U JP 11169186 A	08-04-1999 29-06-1999
WO 9952938 A	21-10-1999	DE 19825585 A DE 19828097 A DE 19831637 A DE 19831639 C AU 4120899 A AU 4481699 A WO 9952515 A AU 4615599 A WO 9966875 A WO 0004031 A WO 0003699 A DE 19923567 A WO 0017233 A	21-10-1999 30-12-1999 27-01-2000 11-05-2000 01-11-1999 01-11-1999 21-10-1999 10-01-2000 29-12-1999 27-01-2000 27-01-2000 06-04-2000 30-03-2000
EP 0256785 A	24-02-1988	DE 3767075 D JP 63152306 A US 4846872 A US 5002602 A	07-02-1991 24-06-1988 11-07-1989 26-03-1991



# PATENT COOPERATION TREATY

From the

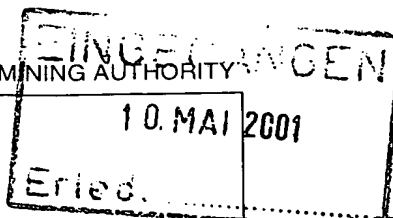
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

by fax and post

To:

Wächtershäuser, Günter  
Tal 29  
D-80331 München  
ALLEMAGNE

# 22 37 59



PCT

## NOTIFICATION OF TRANSMITTAL OF THE INTERNATIONAL PRELIMINARY EXAMINATION REPORT (PCT Rule 71.1)

Date of mailing  
(day/month/year) 08.05.2001

Applicant's or agent's file reference  
EA-PCT-10914

### IMPORTANT NOTIFICATION

International application No.  
PCT/EP99/09870

International filing date (day/month/year)  
13/12/1999

Priority date (day/month/year)  
14/12/1998

Applicant  
BACHER, Adelbert et al

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.
2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.
3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

#### 4. REMINDER

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

Name and mailing address of the IPEA/

European Patent Office  
D-80298 Munich  
Tel. +49 89 2399 - 0 Tx: 523656 epmu d  
Fax: +49 89 2399 - 4465

Authorized officer

Neumann, M

Tel. +49 89 2399-7351



# PATENT COOPERATION TREATY

## PCT

### INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference <b>EA-PCT-10914</b>		<b>FOR FURTHER ACTION</b> See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)	
International application No. <b>PCT/EP99/09870</b>	International filing date (day/month/year) <b>13/12/1999</b>	Priority date (day/month/year) <b>14/12/1998</b>	
International Patent Classification (IPC) or national classification and IPC <b>C12Q1/02</b>			
Applicant <b>BACHER, Adelbert et al</b>			

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.


2. This REPORT consists of a total of 7 sheets, including this cover sheet.

☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

These annexes consist of a total of 6 sheets.

3. This report contains indications relating to the following items:

- I ☒ Basis of the report
- II ☐ Priority
- III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV ☐ Lack of unity of invention
- V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- VI ☐ Certain documents cited
- VII ☒ Certain defects in the international application
- VIII ☒ Certain observations on the international application

Date of submission of the demand  <b>14/07/2000</b>	Date of completion of this report  <b>08.05.2001</b>
Name and mailing address of the international preliminary examining authority:   <b>European Patent Office</b> <b>D-80298 Munich</b> <b>Tel. +49 89 2399 - 0 Tx: 523656 epmu d</b> <b>Fax: +49 89 2399 - 4465</b>	Authorized officer  <b>Linker, W</b>  Telephone No. <b>+49 89 2399 8703</b>



**INTERNATIONAL PRELIMINARY  
EXAMINATION REPORT**

International application No. PCT/EP99/09870

**I. Basis of the report**

1. With regard to the **elements** of the international application (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17)*):

**Description, pages:**

1-41 as originally filed

**Claims, No.:**

1-28 as received on 19/03/2001 with letter of 19/03/2001

**Drawings, sheets:**

1-5 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☐ furnished subsequently to this Authority in written form.
- ☐ furnished subsequently to this Authority in computer readable form.
- ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
- ☐ the claims, Nos.:

# INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/EP99/09870

☐ the drawings, sheets:

5. ☒ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

*(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)*

**see separate sheet**

6. Additional observations, if necessary:

## V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

### 1. Statement

Novelty (N)	Yes:	Claims	1-22
	No:	Claims	23-28
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-28
Industrial applicability (IA)	Yes:	Claims	1-28
	No:	Claims	

### 2. Citations and explanations

**see separate sheet**

## VII. Certain defects in the international application

The following defects in the form or contents of the international application have been noted:

**see separate sheet**

## VIII. Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

**see separate sheet**

**Re Item I**

**Basis of the opinion**

New claims 23-25 have been amended to exclude treatment with fosmidomycin known from document D3. As document D3 is considered relevant not only for novelty but also for inventive step, the use of a disclaimer in this case does not meet the requirements of Article 34 (2) b) PCT. Accordingly, the comments made under item V with respect to claims 23-25 ignore the disclaimer.

**Re Item V**

**Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. Reference is made to the following documents:

D1: LANGE ET AL: 'A family of transketolases that directs isoprenoid biosynthesis via a mevalonate-independent pathway' FASEB JOURNAL,US,FED. OF AMERICAN SOC. FOR EXPERIMENTAL BIOLOGY, BETHESDA, MD, vol. 95, March 1998 (1998-03), pages 2100-2104, XP002116672 ISSN: 0892-6638

⇒ D2: DUVOLD T ET AL: 'Incorporation of 2-C-Methyl-d-erythritol, a Putative Isoprenoid Precursor in the Mevalonate-Independent Pathway, into Ubiquinone and Menaquinone of Escherichia coli' TETRAHEDRON LETTERS,NL,ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, vol. 38, no. 35, 1 September 1997 (1997-09-01), pages 6181-6184, XP004086730 ISSN: 0040-4039 cited in the application

D3: ZEIDLER, JOHANNES ET AL: 'Inhibition of the non - mevalonate 1- deoxy - D- xylulose -5-phosphate pathway of plant isoprenoid biosynthesis by fosmidomycin' Z. NATURFORSCH., C: BIOSCI. (1998), 53(11/12), 980-986 , XP000909111

- 2.1 D1 refers to transketolases which are considered to play a key role in the biosynthesis of plastid-derived isoprenoids essential for growth, development, and defence in plants. On page 2104, left column, second paragraph it was

considered that this pathway provides a unique target for the design of herbicides because it is present in plants but not in animals.

- 2.2 D2 discloses the use of deuterium labelled 2-C-methyl-D-erythritol as a biochemical precursor for generating labelled isoprenoid side chains. On page 6183, last paragraph it was considered that 2-C-methyl-D-erythritol can be converted into an IPP precursor via 1-deoxy-D-xylulose 5-phosphate yielding 2-C-methyl erythritol 4-phosphate.
- 2.3 D3 refers to the use of  $^2\text{H}$ -,  $^{13}\text{C}$ - or  $^{14}\text{C}$ -labelled 1-deoxy-D-xylulose in a study of the non-mevalonate 1-deoxy-D-xylulose-5-phosphate pathway for isoprenoid formation (page 980, right column, second paragraph). In a search for inhibitors of this pathway in plants using 2- $^{14}\text{C}$  pyruvate as a precursor fosmidomycin was found to be effective. Fosmidomycin is a known antibiotic and herbicidal compound.
- 3.1 The subject-matter of claims 23-28 is considered to lack novelty and inventive step according to Article 33(2) and (3) PCT in view of the fact that the herbicidal compound fosmidomycin was already known to inhibit an enzyme in the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate, see abstract of D3 and also EP-A-0 256 785 as cited in the International Search Report. Moreover, page 36, first paragraph of the present specification apparently mentions inhibitors already known in the art.
- 3.2 Many labelled 1-deoxy-D-xylulose/2C-methyl-D-erythritol compounds are known. Moreover, 1-deoxy-D-xylulose 5-phosphate and 2-C-methyl erythritol 4-phosphate were known to be biochemical precursors in the pathway to terpenoids, see D1-D3, in particular D2, page 6183, the paragraph following Figure 3. Therefore, providing alternative labelled compounds according to claims 19-22 does not appear to involve an inventive step according to Article 33(3) PCT.

The claimed compounds have been designed for the method of claim 1, however, claim 1 is also considered to lack an inventive step, see following item 3.3.

- 3.3 The biochemical pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate was

already known to represent a target for herbicides as there was no evidence for its presence in man, animals, fungi and yeasts (see e.g. D1 or the Review by Michel Rohmer as cited in the International Search Report). As furthermore the use of labelled precursors in the study of biosynthetic pathways is common knowledge, see e.g. D3, the skilled person would regard it a normal design procedure to combine all the features set out in claim 1. Thus, the subject-matter of claim 1 does not involve an inventive step and does not satisfy the criterion set forth in Article 33(3) PCT.

The Applicant's argument with respect to inventive step relate to non essential features of claim 1 and can therefore not be accepted for the present scope of the claim, in particular in view of Kobek and Lichtenthaler (1989) as cited in the search report which shows that plastides are commonly used as a test system for herbicides, e.g. page 672, left column relating to chloroplast test systems.

- 3.4 Dependent claims 2-18 relate to normal design options and thus do not appear to contain any additional feature which could be regarded as inventive (Article 33(3) PCT.

#### **Re Item VII**

##### **Certain defects in the international application**

Contrary to the requirements of Rule 5.1(a)(ii) PCT, the relevant background art disclosed in the documents D1 and D3 is not mentioned in the description, nor are these documents identified therein.

#### **Re Item VIII**

##### **Certain observations on the international application**

1. The wording of claim 1, in particular the phrases "biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate", "precursor for generating terpenoids", "product or intermediate downstream from 1-deoxy-D-xylulose 5-phosphate" does not exclude the part of the pathway after IPP formation, which is the same whether or not the starting point is mevalonate (the classical pathway)

or 1-deoxy-D-xylulose 5-phosphate (the pathway referred to in the present application). Since many herbicides are known to inhibit terpenoid biosynthesis, e.g. by inhibition of phytoene desaturase (norflurazon, mentioned on page 39, third paragraph; also diflurazon, norflurazon, or by unknown mechanisms (fluoridone, fluometuron etc.), claim 1 is broader than the description, where only test systems with labelled 1-deoxy-D-xylulose 5-phosphate / 2C-methyl-D-erythritol-4-phosphate are disclosed, contrary to Article 6 PCT.

The Applicant refers to page 37, screening example 4 which, however, only demonstrates that when labelled IPP is used neither F1 nor F2 shows inhibition.

2. The subject-matter of product claims 25-28 lack clarity as the product is only defined in terms of its ability of inhibition in the test system of claim 1, this definition would also comprise any known compounds having this effect. As the specification does not specify any specific structural features of the claimed inhibitors, the aforementioned claims are not supported by the specification and lack clarity, contrary to Article 6 PCT.



## Claims

1. A method for screening for the presence of inhibition of at least one enzyme in the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate in plants comprising the following steps:
  - (a) preparing a suspension of cells or plastids of a plastid-bearing organism in a culture medium for supporting the metabolism of said cells or plastids at least to the extent of said biosynthetic pathway,
  - (b1) adding to said suspension a predetermined amount of a carbon-13-, carbon-14-, deuterium-, or tritium-labelled biochemical precursor for generating terpenoids via said pathway,
  - (c1) incubating the mixture obtained in step (b1) for a predetermined period of time at a predetermined temperature,
  - (d1) separating from said incubated mixture obtained in step (c1) a fraction comprising a product or intermediate downstream from 1-deoxy-D-xylulose 5-phosphate in said pathway,
  - (e1) detecting the concentration of labelled product(s) in said fraction obtained in step (d1),
  - (b2) repeating step (b1) with the addition of a predetermined amount of a chemical test sample under otherwise identical conditions,
  - (c2) to (e2) repeating steps (c1) to (e1) with the mixture obtained in step (b2) under the same conditions as in steps (c1) to (e1) and
  - (f) determining the presence of inhibition of at least one enzyme in said pathway by observation of whether the concentration of labelled product(s) detected in step (e1) is higher than that detected in step (e2).

REPLACEMENT  
ART 30

2. The method according to claim 1, wherein the plastid-bearing organism is a monocotyledonous or dicotyledonous plant.
3. The method according to claim 1, wherein the plastid is a chromoplast or a chloroplast.
4. The method according to claim 1, wherein the biochemical precursor is selected from the group consisting of 1-deoxy-D-xylulose, 1-deoxy-D-xylulose 5-phosphate and 2C-methyl-D-erythritol, 2C-methyl-D-erythritol 4-phosphate and 2C-methyl-D-erythritol 4-pyrophosphate.
5. The method according to claim 1, wherein the culture medium comprises ATP in combination with CTP or a source for CTP.
6. The method according to claim 5, wherein the source for CTP is CMP or CDP.
7. The method according to claim 1, wherein an extraction with a lipophilic organic solvent is used for the separation of terpenoids steps (d1) and (d2).
8. The method according to claim 1, wherein the biochemical precursor is a tritiated biochemical precursor and the product is tritiated water.
9. The method according to claim 8, wherein the separation of tritiated water in step (d1) is effected by a cold trap.
10. The method of claim 9, wherein the cold trap is cooled by liquid nitrogen

REPLACED  
ART 31

or solid carbon dioxide.

11. The method according to claim 8, wherein the separation of a predetermined fraction of water in step (d1) is ascertained by a moisture indicator.
12. The method according to claim 11, wherein the moisture indicator is  $\text{CoCl}_2$ .
13. The method according to claim 8, wherein the concentration of tritiated water is measured by liquid scintillation measurement.
14. The method according to claim 8 characterized in that
  - (a) a multitude of incubations is carried out in parallel in the wells of a first multi-well plate;
  - (b) a second multi-well plate is placed upside down on the first, so that the wells are registered;
  - (c) the second plate is cooled with a coolant while the first plate is preferably heated; and
  - (d) the ice crystals in the wells of the separated and inverted second plate are subjected to liquid scintillation measurement.
15. The method according to claim 14 characterized in that a perforated gasket is placed between the first and the second plate.
16. The method according to claim 14 characterized in that a subset of the wells is used for a moisture indicator.
17. The method according to claim 8 characterized in that the incubation

REPLACED BY  
ART 34 AMDT

medium comprises ATP and/or CTP or a source for CTP.

18. The method according to claim 1 wherein the following additional steps are carried out:
- (g1) adding to the suspension of step (a) of claim 1 a predetermined amount of a carbon-13-, carbon-14-, deuterium- or tritium-labelled isopentenyl pyrophosphate,
  - (c3) to (e3) repeating steps (c1) to (e1) of claim 1 with the mixture obtained in step (g1),
  - (g2) repeating step (g1) with the addition of a predetermined amount of an inhibitor detected in step (f) of claim 1,
  - (h) ascertaining the absence of inhibition of an enzyme in the biosynthetic pathway downstream from isopentenyl pyrophosphate by said inhibitor.
19. A method for inhibiting the growth of a plant by treatment with a herbicidally effective amount of a chemical compound selected from the class of chemical compounds exhibiting inhibition of the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate in the test according to one of claims 1 to 18.
20. A method for inhibiting in chloroplasts of plants an enzyme in the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate by treatment with a chemical compound selected from the class of chemical compounds exhibiting inhibition of the biosynthetic pathway to terpenoids via 1-deoxy-D-xylulose 5-phosphate in the test according to one of claims 1 to 18.

REPLACED BY  
ART 34 AMDT

21. An inhibitor exhibiting inhibition in the test according to one of claims 1 to 17.
22. A herbicidal composition comprising the inhibitor of claim 21 in combination with an agriculturally acceptable carrier.
23. A method for increasing the titer of an intermediate in the biosynthetic terpenoid pathway via 1-deoxy-D-xylulose 5-phosphate upstream from IPP in plants, plant cells, or plastids by treatment with a synthetic inhibitor exhibiting inhibition in the screening test of one of claims 1 to 17.
24. Plants or cells or plastids thereof containing a synthetic inhibitor and exhibiting a titer of an intermediate in the biosynthetic terpenoid pathway via 1-deoxy-D-xylulose 5-phosphate upstream from IPP which is increased over the titer absent the inhibitor.
25. 14-carbon-, 13-carbon-, deuterium- or tritium-labelled 1-deoxy-D-xylulose 5-phosphate or 2C-methyl-D-erythritol 4-phosphate.
26. Tritiated 2C-methyl-D-erythritol 4-phosphate or tritiated 1-deoxy-xylulose 5-phosphate.
27. 2C-Methyl-D-erythritol 4-phosphate tritiated in positions 1 and/or 3 or in the methyl group.
28. 1-Deoxy-D-xylulose 5-phosphate, tritiated in positions 1 and/or 3 and/or 4.

REPLACED BY  
ART 34 AMDT

biosynthetic terpenoid pathway via 1-deoxy-D-xylulose 5-phosphate  
upstream from IPP which is increased over the titer absent the inhibitor.